Progress on the Methods for Preparation of Fish Protein Concentrate from Underutilized Fish Residue: A Short Review

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Abstract. Due to higher cost and existence of various allergens in plant, low-cost proteins derived from animal sources are becoming more attractive to consumers. Currently, there are several methods used for the preparation of fish protein concentrate from underutilized sources. This review elaborates the progress of methods used for the preparation of fish protein concentrate (FPC) from underutilized fish residue. Basically, FPC manufacturing processes involve either the dissolution and precipitation of soluble protein fractions or segregation of undesirable components, namely lipids, water and carbohydrates. The combination of physical and chemical methods has been developed with low investment and operational cost to obtain high product yield and high-quality FPC. In addition, the demand for environmentally friendly process has also encouraged the use of effective and generally regarded as safe solvents. As a conclusion, the preferred FPC preparation method should facilitate the use of green solvents, low investment, energy and operational cost, and the achievement of a high FPC yield with premium product quality.

1 Introduction

Proteins are biopolymeric molecules composed of amino acids in which 20 of the natural ones are linked by amide bonds found in biological chemistry, taking part in almost all cellular activities. Proteins provide pivotal roles in human biochemistry with their major role is to produce the body's building blocks as the precursors of assorted biologically relevant molecules [1]. Hence, either an excess or deficiency of protein can trigger numerous diseases and cause nervous system imperfections, metabolic problems, organ malfunction, and even death. Commonly, sources of dietary protein are considered as either being of animal or plant origin. In fact, proteins of animal origins supply a complete source of protein that contains all essential amino acids, whereas those of plant origins are usually deficient in one or more of the essential amino acids [2]. Meat, egg, milk and fish are the major source of animal-based proteins. Meanwhile, legumes, nuts and mushrooms are the main source of plant-based proteins.

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Based on the fact that fish is the most affordable animal-based proteins, the flesh of freshwater, brackish, and marine fishes can be the most encouraging protein sources for human consumption [3]. Contingent on the species, sex, size, age, culture methods, and the season of the year, fish flesh contains between 15 to 24% protein [4]. Ordinarily, these proteins play pivotal roles to enhance cardiovascular and other related health conditions [5]. Fish proteins display specific functional properties that cause them become extremely good vehicles for the formation of strong gel, stable foams, and stable lipids emulsions with excellent lipids binding and water holding capacity [3, 6]. However, their strong unpleasant fishy odor of these excellent proteins generally brings about unexpected rejection by several groups of people, especially children, elderly, and admitted and non-admitted patients. Therefore, the production of fish protein concentrate (FPC) can be one of the encouraging alternatives to enhance people's acceptance of fish consumption. Basically, the FPC can be defined as a stable fish preparation, in which the protein content is higher than in its fish origin and is intended for human consumption.

Considering the physical and chemical characteristics, the Protein Calorie Advisory Group of the United Nation System has classified FPC into three different types, namely type A, type B and type C. The type-A (white or colorless, no fish smell and taste, contains a minimum of 67% of crude protein, and no higher than 0.75% of fat), the type-B (greyish-brown in color with a strong fish smell and taste due to the presence of about 3% fat) and type-C (a typical fish dinner or crude fish meal provided under acceptable clean conditions) [7]. The FPC usually possesses a smaller particle size than a fish meal with more uniform in both color and texture [8]. The low or negligible oil content in FPC offers more promising consumer acceptance due to the disappearance of the fishy taste in its edible portion.

This review aims to elaborate the progress of FPC manufacturing methods as well as to explain challenges and suggestion of future applications of FPC in food and nutraceutical sectors.

2 Common methods of fish protein concentrate manufacturing

Basically, the aim of fish protein concentrate manufacturing process is to convert whole fish or fish by-products from a highly perishable protein source into a less perishable product. The existing methods applied for the manufacture of FPC involve physical, chemical, and biological treatments [8]. In general, the processes comprise the dissolution of soluble proteins, removal of unwanted fish components, proteins precipitation, drying of the proteins, size reduction of the proteins, and storing the resulted FPC. In brief, the followings are the common methods used for the manufacture of FPC.

2.1 Steaming and aqueous extraction

The simplest method to extract fish protein is carried out by steaming minced fish flesh at 45°C for 25 minutes (Sugiono, 2002) or aqueous solvent extraction, i.e. 0.9% (w/w) sodium chloride, 1 % (v/v) hydrochloric acid with heating (50-60°C) and without heating for 15 minutes, fish/solvent ratio of 1:4 (w/v), precipitation at isoelectric point pH 4.6, filtration or decantation, drying at 50-55°C and ground to achieve yield of 11.6% [9-10].

2.2 Multiple water washing and refining

The raw deboned fish flesh utilized for extracting the myofibrillar proteins has to be fresh and maintained chilled [11]. Primarily, leaching process comprises mixing of minced fish flesh with cold water and water removal by screening using a refiner/strainer and dewatering
using screw press. Generally, number of washing cycles and water volume depend on the fish species and freshness, type of washing unit, and the preferred product quality. However, the process is repeated 2–4 times [12]. The screw press, which usually possess 0.5 mm openings, squeezes water out through compression to remove 82–85 % moisture, which is comparable to that in a fish fillet [11]. Then, the leached fish flesh is mixed with cryoprotecting agents in a silent cutter to produce blocks, packed in plastic bags, and laid on a stainless-steel tray for freezing. The frozen surimi has to be stored at −20°C [13]. However, when oily or dark muscle fish is used as the raw material, such as sardine or mackerel, leaching has to be conducted using alkaline solution to dismiss the undesirable effects of oils and haem proteins [14]. Generally, haem proteins, such as myoglobin and haemoglobin contribute to the red color of dark muscles as well as oil oxidation in the dark muscles that generates unpleasant (rancid) odor [15].

Surimi is minced fish flesh, which contains 16 % water insoluble proteins, 75 % moisture and approximately 9 % freezing stabilizers or cryoprotecting agents [8, 13]. In fact, the insoluble proteins possess elastic texture that permits the formulation of surimi into many types of fish product via advanced processing.

2.3 pH shift method

The minced fish flesh particles are solubilized in five to ten times volume of acid or an alkali solution at approximately pH 2.5 or 11 [16]. After oil and insoluble materials removal via centrifugation, the myofibrillar proteins are precipitated from the solution at their isoelectric point by adjusting the pH, which is around 5.2 to 5.5 [13]. Then, the proteins are sedimented employing a high-speed centrifuge, stabilized from freezing using appropriate cryoprotecting agents and kept frozen prior to further use [17].

2.4 Organic solvents extraction

This process is primarily carried out employing alcohol (ethyl alcohol, isopropyl alcohol, 2-butanol, hexane, ethylene chloride, ethylene dichloride, ethyl acetate, acetone, petroleum ether, or their mixture) solvents to remove water, lipids and fishy tasting substances of raw fish flesh [8]. Basically, solvent selection is based on solvent’s solubilization capacity to dissolve proteins, lipids or water, price and recoverability for further uses [18]. Indeed, the quality of the FPC strongly depends on the raw materials and processing conditions. When FPC is prepared from lean fish, such as hake, its color is light gray to yellowish brown. Meanwhile, the color of FPC prepared from fatty fish species, such as anchovy and herring is usually of a darker gray [19]. Hence, this unpleasant appearance potentially hinders the use of FPC in specific food products [15]. The FPC obtained from this method is stable for 6 months at 5°C when packed in an airtight sealed container. In addition to the common unpleasant appearance, the lack of functional properties is also the main drawback of the FPC manufactured using organic solvent extraction [18, 20].

2.5 Enzymatic or acid hydrolysis

In this method, whole fish or fish by-products are ground and suspended in water, then pH is adjusted prior to the addition of enzyme to the slurry [8, 21]. Hydrolytic enzymes (alcalase) are employed to break the peptide bonds in protein molecules. In order to control the hydrolysis reaction, the raw material is firstly preheated to denature the endogenous proteases. Usually, the reaction occurs within one to a few hours, which depends on the enzyme activity, temperature and other parameters [22]. Because terminating of reaction in an important issue of enzyme catalyzed process, an appropriate sudden change of pH and/or
temperature can be applied to inactivate the enzyme at the end of the batch reaction. After separation of solids, the aqueous layer is clarified, and further concentrated or dried [8, 22].

Generally, FPC obtained from this method usually demonstrates remarkable nutritional properties and biological activities for food and feed [23]. Proteins extracted by partial enzyme hydrolysis are of high quality due to their excellent anti-oxidation properties against peroxidation of both lipids and fatty acids [24]. In fact, the functional properties of FPC obtained from this method strongly depend on the degree of hydrolysis [21].

2.6 Heat treatment

Minced fish flesh is mixed with an equal volume of distilled water (23°C) and homogenized in a high-speed blender for 2 min. The mixture is continuously stirred for 60 min at 85°C to facilitate lipids cells rupture and release them into the liquid phase. Upon centrifugation at 2560 × g for 15 min, the heavier liquid middle layer and insoluble proteins in the semi-solid phase at the bottom are separated from the lipids-rich top layer, collected, and dried by freeze/spray drying and contains 63–81.4% protein. The emulsifying and lipid adsorption capacities of FPC obtained from this method were higher than those of soy protein concentrate [25].

2.7 Combined of the existing methods

This FPC manufacturing processes can involve gamma-irradiation (200 Krad), heat treatment (60°C, 10 min), ultrasonic homogenization with acidification, or microwave irradiation [3, 8]. Although alcalase is the most popular protein degrading enzyme employed for FPC manufacturing, pronase has been reported to solubilize protein more effective in respect to the extent and hydrolysis rate. The FPC exhibits excellent functional properties in terms of emulsifying capacity and water holding capacity [3, 8].

Homogenization of the leached mince fish flesh in three-time volume of water and followed by acidification of the dispersion with 1% acetic acid resulted in a weak gel. After dilution with water and spray drying, a bright yellow powder can be obtained with a faint fishy odor. The resulting FPC contains 5% moisture, 1.2% lipid and 89.5% protein with exceptionally good functional properties. The FPC can be packed in polyester/polyethylene laminate pouch and stored at ambient temperature for 3 months without developing any significant changes of appearance and functional properties.

3 Optimization of manufacturing parameters

The method used and operating condition employed for FPC preparation strongly influence its bioactive properties, which largely depend on the composition and molecular weight of the amino acids. In general, the functional properties demonstrated by the FPC are attributed by the presence of particular protein fragments encrypted within the sequence of the parent protein and possess the targeted active functional sites, which are developed during processing and/or digestion. The surface hydrophobicity of the amino acids present in a protein is the principal parameter that strongly determines their functional properties [26]. Hence, the composition of hydrophobic amino acids presents in the FPC become the key parameters for predicting the quality, conversion factor, and indirect functional properties of the FPC, which exhibit numerous biological and physiological activities of the human body and in keeping healthy [27].
4 Challenges of FPC manufacturing process development

Some challenges exist within FPC processing methods and applications. An extensive cost-benefit appraisal of fish proteins in comparison with soy proteins has been studied with respects to some hypotheses and important facts [28], which include: the raw material and manufacturing costs to produce fish proteins are roughly ten times higher than that of soy proteins, the method used and operating conditions employed for FPC preparation strongly determines its physicochemical and functional properties that directly depend on the composition and molecular weight of its amino acids, the global demand for the use of green, non-toxic and efficient solvents, low total energy requirement for protein extraction and drying, simple and economical operational process [3, 8, 28]. Further advancement of technology for fish proteins extraction, concentration, drying, formulation and applications in consumer products requires more complete research on addressing the hypotheses. FPC drying using spray drying, freeze drying, vacuum oven drying or rotary drying are good examples of total energy requirement.

The global demand for FPC with acceptable physicochemical and functional properties is another challenge in FPC manufacturing.

5 Future promising applications of FPC

To fulfill the growing requirement of high-quality FPC and any food and nutraceutical products containing fish proteins, it is of large significance to use fish as raw materials more efficiently and to look for new resources of fish. Processing methods, which can employ low-cost raw material and whole fish instead of fillets would build a remarkable economic benefit to the fish protein raw material industry, as new sources of fish inappropriate for conventional processes could be received by the market, and their production could be improved to fulfill the global requirement. Refer to the aforementioned FPC processing methods, the pH-shift processing appears to possess the highest promise on by-products and underutilized fish species. However, to enhance the accomplishment of this method and its respective products, it is crucial to conduct more advanced research to drive the commercial applications of FPC either as a food ingredient or as food products.

Extensive research should be concentrated on the usage of FPC for human consumption in lieu animal consumption, even though the latter cannot be disregarded. One of the promising food applications to be viewed is the utilization of FPC as a water-binder in seafood processing. Considering that FPC can strongly compete with and even surpass phosphates as water-binders, it could bring a noteworthy role for the seafood processing sector. Future research should also be addressed towards finding efficient methods to efficiently and economically stabilize the proteins from functional and oxidative (e.g., lipid oxidation) changes during processing and storage. Because the future for functional FPC is encouraging, the correct perspective from consumer, industry, government and academia great progress can be built in the near future.

6 Conclusion

Fish is abundantly available at low cost and considered as the safest and most ideal protein source for human consumption, nonetheless, its strong unpleasant fishy odor generally induce unexpected rejection by some groups of people. Transformation of fish flesh into FPC can be an encouraging alternative to preserve the nutrients and enhance consumer acceptance. FPC can be produced from underutilized fish and fish by-products and used for supplementing children’s and patient’s diets, especially regions with are lacking of consuming fresh or
frozen fish. The production cost depends on the raw material and the process used, whereas the market depends on the price and functional properties of FPC. FPC can be successfully marketed in the regions where fish powder is consumed with the staple dish. The pH shift method demonstrates its unrivaled performance in terms of product yield and nutrient composition compared to other processes for the preparation of FPC. The solvent employed for FPC manufacture should be a mixture of polar and non-polar green solvents with the composition largely depends on the lipid composition and water content of the fish utilized as the raw material. In fact, the fish species, processing method, and operating conditions firmly decide the functional properties of the resulting FPC. Based on its nutrient composition, FPC offers encouraging potentials as a versatile fortifying food supplement for the market. The pH shift on the lipid composition and water content of the fish utilized in various cosmetic and health care applications. Moreover, FPC has also broadly been marketed in the regions where fish powder is consumed with the staple dish. The pH shift on the lipid composition and water content of the fish utilized

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